

APPENDIX VI. ANALYTICAL METHODS, 2002 FSIS NATIONAL RESIDUE PROGRAM

Implementing the NRP requires analytical methods for detecting, quantifying, and identifying residues that may be present in meat, poultry, and egg products. Results from the residue testing are used by the Agency to determine whether a product is adulterated. The following Table AVI, *Analytical Methods*, describes the types of methods used by FSIS to conduct analyses.

KEY TO ABBREVIATIONS

APCI -- Atmospheric Pressure Chemical Ionization

Confirm. -- Confirmatory Method

Determ. -- Determinative Method

ECD -- Electron Capture Detector

ELISA -- Enzyme-Linked Immuno Sorbent Assay

GC -- Gas Chromatograph

GPC -- Gel Permeation Chromatography

HPLC -- High Performance Liquid Chromatography

Method Detection Limit -- The lowest amount of individual residue or sample component that can be reliably observed or found in the sample matrix by the current appropriate analytical methodology.

Minimum Reportable Level -- Lowest level at which an analytical result is reported.

MS -- Mass Spectrometry

NA -- Not Applicable

ppb -- Parts per billion

ppm -- Parts per million

SIM -- Selected-Ion Monitoring Mode

TBD -- To Be Determined

Table AVI
Analytical Methods^a
2002 National Residue Program

| Compound Class | Compound | Analytical Method | | | Minimum Proficiency Level ^a | | |
|------------------------------|---------------------|-------------------|------------------------------|-------------------------------|--|------------------------------|--------------------------------------|
| | | Screen | Determinative (quantitative) | Confirmatory (identification) | Screen | Determinative (quantitative) | Confirmatory (identification) |
| Antibiotics | Carbadox | | GC-ECD | TBD | | 15 ppb | TBD |
| | Chloramphenicol | | GC | GC-MS | | 0.30 ppb | 0.30 ppb |
| | Florfenicol | | HPLC | GC-MS | | 1.9 ppm (L) | 1.9 ppm (L) |
| Antibiotics : beta-Lactams | Penicillin | 7-Plate Bioassay | Bioassay | | | 0.01 ppm | |
| Antibiotics : Tetracyclines | Chlortetracycline | 7-Plate Bioassay | Bioassay | HPLC (chemistry) | 0.5 ppm | 0.08 ppm | |
| | Oxytetracycline | | | | | | |
| | Tetracycline | | | | | | |
| Antibiotics: Macrolides | Clindamycin | 7-Plate Bioassay | | MS | | | 0.1 ppm |
| | Erythromycin | | Bioassay | | | 0.05 ppm | 0.1 ppm |
| | Lincomycin | | | | | | 0.1 ppm |
| | Pirlimycin | | | | | | 0.1 ppm |
| | Tilmicosin | | HPLC- Ion Pairing | | | 300 ppb (M) 600 ppb (L,K) | 600 ppb |
| | Tylosin | | Bioassay | | | 0.2 ppm | 0.1 ppm |
| Antibiotics: Aminoglycosides | Amikacin | 7-Plate Bioassay | | MS | | | 1.0 ppm (L,K), 0.4 ppm (M) |
| | Apramycin | | | | | | 0.4 ppm (K) 0.1 ppm (L,M) |
| | Dihydrostreptomycin | | Bioassay | | | | 0.4 ppm (L,K,M) |
| | Gentamicin | | Bioassay | | | 0.15 ppm | 0.1 ppm (K,M) |
| | Hygromycin | | | | | | 1.0 ppm (L,K) 0.4 ppm (M) |
| | Kanamycin | | | | | | 4.0 ppm(L), 2.0 ppm (K), 0.4 ppm (M) |
| | Neomycin | | Bioassay | | | 0.25 ppm | 0.1ppm (K,M) |
| | Spectinomycin | | | | | 10.0 ppm | 1.0 ppm (L) 0.4 ppm (K) 0.25 ppm (M) |
| | Streptomycin | | Bioassay | | | 0.1 ppm | 0.4 ppm (L,K,M) |
| | Tobramycin | | | | | | 1.0 ppm (L) 0.1 ppm (K,M) |

Table AVI – *continued*
Analytical Methods^a
2002 National Residue Program

| Compound Class | Compound | Analytical Method | | | Minimum Proficiency Level ^a | | |
|---|---------------------------------------|-------------------|---------------------------------|----------------------------------|--|---------------------------------|----------------------------------|
| | | Screen | Determinative (quantitative) | Confirmatory (identification) | Screen | Determinative (quantitative) | Confirmatory (identification) |
| Arsenicals | Arsenicals | | AA | AA | | 0.2 ppm | 0.2 ppm |
| Avermectins | Ivermectin, Doramectin, Moxidectin | | HPLC | APCI/LC/MS | | 7.5 ppb | 25 ppb |
| beta -Agonists | Cimaterol | ELISA | | | 6 ppb | | |
| | Clenbuterol | ELISA | | LC-MS-MS | 3 ppb | | TBD |
| | Ractopamine | | HPLC | LC/MS | | 1 ppb | 1 ppb |
| | Salbutamol | ELISA | | | 3 ppb | | |
| Hormones, synthetic | DES | | GC-MS | GC-MS | | 0.5 ppb | 1.0 ppb |
| | Zeranol | | GC-MS | GC-MS | | 0.5 ppb | 1.0 ppb |
| Nonsteroidal Anti- inflammatory Drugs (NSAIDs) | Phenylbutazone | ELISA | | LC-MS-MS | 50 ppb | | 50 ppb |
| | Flunixin | ELISA | HPLC | LC/MS | 50 ppb | 31.3 ppb | 125 ppb |
| Steroids | Melengesterol Acetate | | GC | LC/MS | | 5 ppb | 12.5 ppb |
| Sulfonamides | Sulfapyridine | TLC | GC-MS | | 0.05 ppm | 0.05 ppm | |
| | Sulfadiazine | | | | | | |
| | Sulfathiazole | | | | | | |
| | Sulfamerazine | | | | | | |
| | Sulfamethazine | | | | | | |
| | Sulfachloropyridazine | | | | | | |
| | Sulfamethoxyprydazine | | | | | | |
| | Sulfaquinoxaline | | | | | | |
| | Sulfadimethoxine | | | | | | |
| | Sulfaethoxypyridazine | | | | | | |
| | Sulfaphenazole | | | | | | |

Table AVI – *continued*
Analytical Methods^a
2002 National Residue Program

| Compound Class | Compound | Analytical Method | | | Minimum Proficiency Level ^a | | |
|-----------------------------|-----------------------|-------------------|---------------------------------|----------------------------------|--|---------------------------------|----------------------------------|
| | | Screen | Determinative (quantitative) | Confirmatory (identification) | Screen | Determinative (quantitative) | Confirmatory (identification) |
| Sulfonamides (continued) | Sulfatroxazole | | TLC | GC-MS | | 0.05 ppm | 0.05 ppm |
| | Sulfisoxazole | | | | | | |
| | Sulfadoxine | | | | | | |
| CHCs/COPs/PCBs | Aldrin | | GPC with GC-EC | GC-MS | | 0.10 ppm | |
| | <i>alpha</i> -BHC | | | | | 0.10 ppm | 0.01 ppm |
| | Captan | | | | 0.04 ppm | | |
| | Carbophenothion | | | | 0.06 ppm | | |
| | Chlorfenvinphos | | | | | 0.06 ppm | |
| | Chlorpyrifos | | | | | 0.10 ppm | |
| | <i>cis</i> -chlordane | | | | | 0.30 ppm | |
| | Coumaphos-O | | | | | 0.20 ppm | |
| | Coumaphos-S | | | | | 0.20 ppm | |
| | Dieldrin | | | | | 0.10 ppm | 0.01 ppm |
| | Endosulfan I | | | | 0.01 ppm | | |
| | Endosulfan II | | | | | 0.06 ppm | |
| | Endrin | | | | | 0.10 ppm | 0.03 ppm |
| | HCB | | | | | 0.10 ppm | 0.01 ppm |
| | Heptachlor epoxide | | | | | 0.10 ppm | 0.10 ppm |
| | Heptachol | | | | | 0.10 ppm | 0.01 ppm |
| | Kepone | | | | 0.06 ppm | | |
| | Lindane | | | | | 0.10 ppm | 0.01 ppm |
| | Linuron | | | | 0.50 ppm | | |
| | Methoxychlor | | | | | 0.50 ppm | 0.15 ppm |
| | Mirex | | | | | 0.10 ppm | |

Table AVI – *continued*
Analytical Methods^a
2002 National Residue Program

| Compound Class | Compound | Analytical Method | | | Minimum Proficiency Level ^a | | |
|-------------------------------|-------------------------|-------------------|---------------------------------|----------------------------------|--|---------------------------------|----------------------------------|
| | | Screen | Determinative (quantitative) | Confirmatory (identification) | Screen | Determinative (quantitative) | Confirmatory (identification) |
| CHCs/COPs/PCBs (continued) | Nonchlor | | GPC with GC-EC | GC-MS | | 0.15 ppm | |
| | o,p'-DDT | | | | 0.15 ppm | | |
| | Oxychlordane | | | | | 0.06 ppm | 0.1 ppm |
| | p,p'-DDE | | | | | 0.10 ppm | 0.02 ppm |
| | p,p'-DDT | | | | | 0.15 ppm | 0.04 ppm |
| | p,p'-TDE | | | | | 0.15 ppm | 0.04 ppm |
| | PCB 1260 | | | | | 0.50 ppm | |
| | PCB 1254 | | | | | 0.50 ppm | |
| | Phosalone | | | | 0.02 ppm | | |
| | Ronnel | | | | | 0.06 ppm | |
| | Stirofos | | | | | 0.06 ppm | |
| | Toxaphene | | | | | 1.00 ppm | |
| | <i>trans</i> -chlordane | | | | | 0.30 ppm | |

a Laboratory analytical methods reported here are current through May of 2004.

b Minimum Proficiency Level: The lowest amount of individual residue or sample component that FSIS requires its laboratories to reliably detect, quantify, or confirm. This is usually the lowest amount for which the method used by FSIS laboratories has been validated.

Key:

L = Liver

K = Kidney

M = Muscle

AA = Atomic Absorption Spectroscopy

CHCs = Chlorinated hydrocarbons

COPs = Chlorinated organophosphates

PCBs = Polychlorinated biphenyls

GC = Gas Chromatography

MS = Mass Spectroscopy

GPC = Gel Permeation Chromatography

Table AVI – *continued*
Analytical Methods^a
2002 National Residue Program

TLC = Thin Layer Chromatography

ECD = Electron Capture Detection

ELISA = Enzyme Linked Immunosorbent Assay

ppm = parts per million

ppb = parts per billion

APCI = Atmospheric Pressure Chemical Ionization

HPLC = High Performance Liquid Chromatography

TBD = To be determined

APPENDIX VII. STATISTICAL TABLE

Table VII, *Statistical Table*, indicates the number of samples required to ensure detection of a violation that affects a given percentage of the sampled population.

**Table AVII
Statistical Table**

| Percentage Violative in Sampled Population | Probability of Detection (Percent) | | | |
|---|------------------------------------|-------|-------|--------|
| | 90 | 95 | 99 | 99.9 |
| | Samples Required | | | |
| 10 | 22 | 29 | 44 | 66 |
| 5 | 45 | 59 | 90 | 135 |
| 1 | 230 | 299 | 459 | 688 |
| 0.5 | 460 | 598 | 919 | 1,379 |
| 0.1 | 2,302 | 2,995 | 4,603 | 6,905 |
| 0.05 | 4,605 | 5,990 | 9,209 | 13,813 |